

# Document made available under the Patent Cooperation Treaty (PCT)

International application number: PCT/NL05/000147

International filing date: 01 March 2005 (01.03.2005)

Document type: Certified copy of priority document

Document details: Country/Office: EP  
Number: 04075638.9  
Filing date: 01 March 2004 (01.03.2004)

Date of receipt at the International Bureau: 03 May 2005 (03.05.2005)

Remark: Priority document submitted or transmitted to the International Bureau in compliance with Rule 17.1(a) or (b)



World Intellectual Property Organization (WIPO) - Geneva, Switzerland  
Organisation Mondiale de la Propriété Intellectuelle (OMPI) - Genève, Suisse



Europäisches  
Patentamt

European  
Patent Office

Office européen  
des brevets

3 1. 03. 05

Bescheinigung

Certificate

Attestation

Die angehefteten Unterlagen stimmen mit der ursprünglich eingereichten Fassung der auf dem nächsten Blatt bezeichneten europäischen Patentanmeldung überein.

The attached documents are exact copies of the European patent application described on the following page, as originally filed.

Les documents fixés à cette attestation sont conformes à la version initialement déposée de la demande de brevet européen spécifiée à la page suivante.

Patentanmeldung Nr. Patent application No. Demande de brevet n°

04075638.9

Der Präsident des Europäischen Patentamts;  
Im Auftrag

For the President of the European Patent Office

Le Président de l'Office européen des brevets  
p.o.

R C van Dijk





Anmeldung Nr:  
Application no.: 04075638.9  
Demande no:

Anmeldetag:  
Date of filing: 01.03.04  
Date de dépôt:

Anmelder/Applicant(s)/Demandeur(s):

Nederlandse Organisatie voor Toegepast-  
Natuurwetenschappelijk Onderzoek TNO  
Schoemakerstraat 97  
2628 VK Delft  
PAYS-BAS

Bezeichnung der Erfindung/Title of the invention/Titre de l'invention:  
(Falls die Bezeichnung der Erfindung nicht angegeben ist, siehe Beschreibung.  
If no title is shown please refer to the description.  
Si aucun titre n'est indiqué se référer à la description.)

Process for the production and recovery of hydrocarbons

In Anspruch genommene Priorität(en) / Priority(ies) claimed / Priorité(s)  
revendiquée(s)  
Staat/Tag/Aktenzeichen/State/Date/File no./Pays/Date/Numéro de dépôt:

Internationale Patentklassifikation/International Patent Classification/  
Classification internationale des brevets:

C12P/

Am Anmeldetag benannte Vertragstaaten/Contracting states designated at date of  
filing/Etats contractants désignées lors du dépôt:

AT BE BG CH CY CZ DE DK EE ES FI FR GB GR HU IE IT LU MC NL  
PL PT RO SE SI SK TR LI



P67968EP00

Title: Process for the production and recovery of hydrocarbons

(68)

The invention is directed to a process for the production and recovery of chemicals, in particular hydrocarbons, from a fermentation medium.

Production of hydrocarbon starting materials by fermentation is an alternative to production from petroleum. Fermentation uses renewable resources and creates much less hazardous waste. The fermentative production of a few chemicals has been reported. Unfortunately, this is not a commercially feasible alternative for a great many desirable compounds because the compound of interest is produced in small amounts by the biocatalytic organisms. Even organisms genetically engineered to produce enhanced amounts of the desired compound often do not produce concentrations of the product great enough to justify the investments necessary to develop a commercial biofermentation process. This can be particularly true when the product is toxic to the cells or is regulated by a negative feedback mechanism, thereby limiting the potential concentration of the product in the fermentation medium.

WO-A-00/18942 describes a process wherein 4-hydroxybenzoic acid (PHB) is produced by fermentation. According to this process, the product PHB is removed from the fermentation during at least a portion of the fermentation by passing the fermentation medium through an anion exchange resin which binds PHB. Subsequently the anion exchange resin is extracted to remove bound PHB. Although this known process is said to result in improvements in recovery of the product and higher production of the product by the biocatalyst, there remain a number of disadvantages. First, the ion exchange resin needs to be applied in the form of a packed bed, which may result in considerable pressure drop and fouling of the bed. Moreover, ion exchange resins may have a negative influence on the biocatalyst (*i.e.*, the microorganisms), since these resins can be poisonous to these biocatalysts. Amongst others for this reason, the extraction of product from the medium is carried out by contacting the

medium in-stream (*viz.* outside the fermentor) with the resin, so that contact between resin and biocatalyst is minimized. Furthermore, because of fouling of the resin, amongst others, it is not possible to carry out this process in a continuous fashion. For these reasons, the product yield of the process of  
5 WO-A-00/18942, like the product yield of other known processes, is limited.

Furthermore, WO-A-00/73485 describes a process for extraction of fermentation product from a broth, using a solvent that is encapsulated in a membrane. However, this process, as well as other membrane based processes for the recovery of products from fermentation processes, are hampered by  
10 slow absorption kinetics, as a result of the high mass transfer barrier posed by the membrane. In addition, regeneration of these capsule absorbentia is generally difficult.

Alternatively, liquid-liquid extraction by directly contacting the broth with a solvent for the product generally is also not an option, since this  
15 generally does not result in the desired suspended droplets of solvents, which subsequently coalesce so that they could be separated by gravity, but rather this leads to the formation of an emulsion. These emulsions may be very stable and consequently it is very difficult or impossible to obtain the product from the solvent.

It is an object of the present invention to provide a process for the  
20 production of hydrocarbons, such as 4-hydroxybenzoic acid and benzaldehyde, which process, at least in part, overcomes the above-mentioned disadvantages. Other examples of hydrocarbons that may be produced according to the present invention are catechols (*e.g.* 3-methylcatechol), benzylalcohol, cinnamic  
25 acid, as well as mixtures of these and other hydrocarbons.

It has been found that by using solvent impregnated carrier particles to perform the separation of the product from the broth in the fermentation process, this object can be met.

Thus, in one aspect the present invention is directed to a process for the production of hydrocarbon from a fermentation liquid comprising the steps of:

- 5    - forming said hydrocarbon from said fermentation liquid using a biocatalyst;
- contacting said fermentation liquid with a solvent impregnated carrier, whereby said formed hydrocarbon is sorbed by said solvent impregnated carrier;
- 10   - regenerating said solvent impregnated carrier, whereby a stream comprising said hydrocarbon is obtained; and
- optionally, recycling said regenerated solvent impregnated carrier.

The term "sorption" as used herein, covers absorption, adsorption as well as combinations thereof. This term is also intended to cover absorption or adsorption as a result of chemical reaction of the product component in the solvent, which may be assisted by including additives in the solvent, so as to  
15   facilitate reactive extraction.

The solvent impregnated carrier (by the present inventors sometimes also referred to as "SISCA", which is an acronym for "solvent impregnated smart carrier") that is used according to the present invention  
20   comprises a porous carrier and a solvent immobilized in the pores of said porous carrier. The carriers are suitably in the form of small particles, typically having a diameter of several tenths of mm to several mm, *e.g.* 0.3 mm to 20 mm, preferably 0.4 mm to 5 mm. It was found that for these dimensions an optimal balance is obtained between mass transfer properties on the one  
25   hand and ease of separation of the SISCA containing the product from the fermentation broth on the other hand. The carriers can be made of any material that is non- biodegradable and has a binding with the solvent that is sufficiently strong to immobilize the solvent. The carrier material is hydrophobic. It must furthermore be able to withstand the regeneration  
30   conditions, *e.g.* steam of high temperature, when regeneration is carried out



with steam. Also, to avoid infection of the fermentor with foreign microorganisms, the carrier material must be such that it can be sterilised without degrading.

Preferably the carrier comprises a polymeric material, although  
5 certain types of ceramic materials such as silica, alumina or aluminosilicates (e.g. zeolites) may be used as well. Preferably the carrier comprises a crosslinked polymeric material, e.g. polymers crosslinked with divinylbenzene (DVB). Preferred polymers are polystyrene and polypropylene, optionally crosslinked. Other suitable polymeric materials are polymeric resins such as  
10 Amberlite™ resins, e.g. XAD4, which is a hydrophobic polymer of styrene and DVB; or XAD16, which is a hydrophobic polymer of DVB. Typically, the carrier is obtained by extrusion of the (polymeric) material.

The pores in the carrier particles have dimensions that preferably range from 0.05 to 100  $\mu\text{m}$ , more preferably from 0.1  $\mu\text{m}$  to 10  $\mu\text{m}$ . The porosity  
15 of the carrier particles is preferably from ca. 65 to ca. 85 vol.%, more preferably from 70 to 80 vol.%.

Suitable solvents for use in the present invention are those that display sufficient bonding with the carrier, *viz.* the solvent must have a suitable surface tension. Furthermore, the solvent should have a sufficient  
20 affinity for the hydrocarbon to be recovered from the medium, *viz.* the distribution coefficient should be favourable. The solvent should have a water solubility that is sufficiently low to avoid losses of solvent when in contact with the medium. The solvent should not be too volatile, so as to avoid evaporation as much as possible. It should not be explosive. Also it should essentially not  
25 be biodegradable under the conditions employed. The solvent must be able to withstand the regeneration step. Moreover, it should preferably be possible to sterilize the solvent if this is required.

Suitable solvents include di-isodecylphtalate (DIDP), castor oil, octanol, decanol, cumene, petroleum ether and the like.

Suitable microorganisms (biocatalysts) for use in the present invention are one or more of *Pseudomonas putida*, *Escherichia coli*, *Sacharomyces cerevisiae*, *Lactobacillus* species, *Aspergillus niger*, and the like.

One of the advantages of the present invention is that it provides  
5 integration of production by biochemical reaction and separation. Thus these processes may be combined in a simple unit operation, which reduces installation and operating costs considerably. In this way, *in situ* sorption of the products takes place, which ensures favourable reaction conditions, since product inhibition of the biocatalyst no longer occurs or occurs to a more  
10 limited extent.

The solvent impregnated carriers used in the present invention may be prepared by methods known in the art. For instance, polymers may be impregnated with the solvent by simply contacting the carrier polymer with a mixture of the solvent and a second, more volatile solvent. After impregnation,  
15 the more volatile solvent is evaporated under reduced pressure, by which the solvent impregnated carrier is obtained. It is also possible to melt a polymer carrier at a high temperature and subsequently mix the molten polymer with the solvent and optional carriers. During cooling, a porous polymeric matrix is formed, the pores of which are filled with the solvent, thus forming the solvent  
20 impregnated particles.

According to the present invention the hydrocarbon when it is formed is absorbed and/or adsorbed from the broth by the solvent impregnated carrier. This means that the hydrocarbon can be dissolved in the solvent, which is comprised by the solvent impregnated carrier, it may adsorb on the  
25 surface of the carrier particles, or both. In addition to dissolving in the solvent, it is also possible that the hydrocarbon undergoes a chemical reaction in or with the solvent, by which sorption of the hydrocarbon from the medium is improved. In order to improve chemical reaction, certain agents may be present in the solvent. These agents should be mechanically and biologically  
30 stable, must have a strong but reversible interaction with the hydrocarbon,

must be selective for the hydrocarbon to be produced as compared to other components, must be essentially insoluble in water and soluble in the extraction solvent, must provide for sufficiently fast reaction kinetics, and must be able to withstand the regeneration conditions. In general the same requirements given for the solvent hereinabove apply to these agents.

5 Examples of such agents are complexing agents, such as amines, *e.g.* tri-octylamine, di-ethylamine, tri-ethylamine; or certain organo-phosphorus compounds such as DEHPA (di-(2-ethylhexyl)phosphoric acid), DEHTPA (di(2-ethylhexyl)dithiophosphoric acid), HEHEHP (2-ethylhexyl-phosphonic acid

10 mono-2-ethylhexyl ester).

The contacting of the fermentation liquid with the solvent impregnated carrier can be effected in any known manner. In one embodiment, a configuration is used wherein the particles are fed at the bottom of the fermentor and are allowed to rise due to the density difference of the particles

15 and the fermentation liquid. Thus, the density of the solvent impregnated particles (solvent + carrier) in this embodiment, is preferably less than the density of water. More preferably the density of the solvent impregnated particles is about 900 kg/m<sup>3</sup> or less.

The regeneration of the solvent impregnated carrier may be carried

20 out by using steam, by back-extraction, by heating, or by combinations thereof. In one embodiment the regeneration is carried out using steam, by contacting the loaded carrier with *e.g.* steam having a temperature of 100 to 200 °C and a pressure of 1 to 20 bara.

Back-extraction involves contacting the loaded solvent impregnated

25 carrier with a further solvent that has a higher affinity for the product than the solvent. If the product is chemically bonded by the additives present in the solvent or carrier, it can also be recovered by allowing the reverse reaction to take place, by which the product is released. For example, in the case of the back extraction of organic acids, alkaline solutions can be used.

Fig. 1 schematically shows a suitable configuration for carrying out the present invention. According to the embodiment depicted in Fig. 1, to fermentor (1) starting material (2) is fed, *e.g.* continuously. The starting material is converted to the hydrocarbon by the microorganisms in the fermentor. The fermentor can *e.g.* be a CSTR or a fluidized bed type reactor. It is also possible to operate the process intermittently, *e.g.* as a fed-batch. The solvent impregnated particles are fed to the fermentor at (6). In the fermentor (1) the solvent impregnated carriers, which are selective for the product hydrocarbon, become loaded with the product hydrocarbon. Subsequently the loaded carriers are recovered, preferably at the top of the fermentor. The carriers loaded with the product hydrocarbon (3) are then fed to a regeneration section. This may be in the form of a steam stripper (4), in which steam (5) is contacted with the loaded carriers. Operating conditions of the stripping section include a steam pressure of up to 20 bara. During stripping, the hydrocarbon evaporates from the solvent in the solvent impregnated carriers. The solvent itself remains in the carriers. A gaseous product stream leaves the stripping section, which after cooling at (7) is separated at (8) in an aqueous fraction (not shown) and the product stream (9). In the embodiment of Fig. 1, the solvent impregnated carriers are fed at (or near) the bottom of the fermentor. By selecting solvent impregnated carriers having a density that is lower than that of the broth in the fermentor (*viz.* a density typically lower than that of water), the carriers are allowed to travel upward and to become loaded with product. At (or near) the top of the fermentor the carriers can be collected very easily by simply skimming the carriers off.

Another option is to use solvent impregnated carriers with a higher density than the density of water. In that case the unloaded carriers are fed at or near the top of the fermentor and the loaded carriers are recovered at the bottom of the fermentor.

Apart from rising or sinking solvent impregnated carriers, other contacting processes for capturing components from the broth can be used, such as packed bed or expanded bed contacting steps.

5

10

Claims

1. Process for the production of hydrocarbon from a fermentation liquid comprising:
  - forming said hydrocarbon from said fermentation liquid using a biocatalyst;
  - contacting said fermentation liquid with a solvent impregnated carrier,
  - 5 whereby said formed hydrocarbon is sorbed by said solvent impregnated carrier;
  - regenerating said solvent impregnated carrier, whereby a stream of said hydrocarbon is obtained; and
  - optionally, recycling said regenerated solvent impregnated carrier.
- 10 2. Process according to claim 1, wherein said solvent impregnated carrier comprises a polymeric carrier.
3. Process according to claim 2, wherein said polymeric carrier comprises one or more components selected from polystyrene, polypropylene, polystyrene, polytetrafluoroethylene, silicone, polyethylene, and (regenerated)
- 15 cellulose, which are optionally crosslinked.
4. Process according to claim 1, wherein said solvent impregnated carrier comprises an inorganic carrier, preferably selected from silica, alumina, aluminosilicates and combinations thereof.
5. Process according to any of the previous claims, wherein said
- 20 regeneration is carried out by using steam, by back-extraction, by heating, or by combinations thereof.
6. Process according to any of the previous claims, wherein said product hydrocarbon is 4-hydroxybenzoic acid, benzaldehyde, or a mixture thereof.
- 25 7. Process according to any of the previous claims, wherein said biocatalyst is selected from *Pseudomonas putida*, *Escherichia coli*, *Sacharomyces cerevisiae*, *Lactobacillus* species, *Aspergillus niger*.

8. Process according to any of the previous claims, wherein said solvent impregnated carriers are inserted essentially at or near the bottom of a fermentor containing said fermentation liquid and are collected essentially at or near the top of said fermentor, wherein said solvent impregnated carriers
- 5 have a density that is lower than that of said fermentation liquid.
9. Process according to any of the previous claims, which is carried out continuously.

Title: Process for the production and recovery of hydrocarbons

Abstract

The invention is directed to a process for the production and recovery of chemicals, in particular hydrocarbons, from a fermentation medium, wherein solvent impregnated carriers are used. Accordingly, the invention provides a process for the production of hydrocarbon from a fermentation liquid comprising:

- forming said hydrocarbon from said fermentation liquid using a biocatalyst;
- contacting said fermentation liquid with a solvent impregnated carrier, whereby said formed hydrocarbon is sorbed by said solvent impregnated carrier;
- regenerating said solvent impregnated carrier, whereby a stream of said hydrocarbon is obtained; and
- optionally, recycling said regenerated solvent impregnated carrier.





1/1

68

Fig. 1



